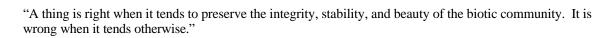
BENCHMARK BIOLOGY OF MONTANA REFERENCE STREAMS

by

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Aldo Leopold *Land Ethic*<u>A Sand County Almanac</u>

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SUMMARY

This report describes the composition and structure of benthic macroinvertebrate (mostly insect) and periphyton (algae) communities inhabiting select least-impaired reference streams in Montana's six major ecoregions in the summer of 1990. The report also describes the fish fauna of these streams and provides supporting information on water quality, macroinvertebrate habitat, and overall stream condition.

Periphyton communities in mountain streams tend to be dominated by blue-green algae and diatoms while those in plains streams tend to be dominated by green algae and diatoms. The number of periphyton taxa tends to be larger in plains streams than in mountain streams. Plains streams support more pollution-tolerant diatoms than mountain streams. The percent relative abundance of diatoms in the genera *Navicula* and *Nitzschia* appears to be a good indicator of sedimentation. We recommend this metric, along with diatom species diversity (Shannon) and an index derived from Lange-Bertalot's pollution tolerance groups. Periphyton metrics appear to partition best into mountain and plains regions, with foothill streams included in the mountain region.

DECORANA suggests that Montana macroinvertebrate communities partition best into mountain, plains, and foothill communities, but not by the ecoregions of Omernik and Gallant (1987). The number of macroinvertebrate taxa tends to be larger in mountain streams than in plains streams, reflecting the larger number of mayfly, stonefly, and caddisfly (EPT) taxa in the mountain streams. Snails, crustaceans, and non-EPT taxa dominate the macroinvertebrate communities of plains streams, while foothill streams have an intermediate fauna. Several macroinvertebrate metrics are proposed. These employ individual taxa or assemblages of taxa that are ubquitous within a region and are strongly correlated with water quality. Macroinvertebrate habitat assessments used for this study were a weak predictor of macroinvertebrate community health.

Reference streams in the mountain and foothill ecoregions supported cold-water fish species, while reference streams in the plains ecoregions supported cool-water and warm-water species. There is more uniformity in the makeup of fish faunas among plains reference streams than there is among mountain reference streams. Mountain and foothill streams have depauperate fish faunas compared to plains streams.

Based on streamflow estimates and water quality measurements, reference streams tend to cluster into a mountain group and a plains group. Foothills streams have intermediate values that are closer to those for mountain streams. Mountain and foothill streams tend to be larger, colder, fresher, clearer, softer, and less alkaline than plains streams. Mountain and foothill streams have calcium bicarbonate waters; plains streams have sodium sulfate or sodium bicarbonate waters. Mountain streams tend to be phosphorus limited; plains streams tend to be nitrogen limited.

Over half of the selected reference streams are impaired, and most of these are in the plains ecoregions. Despite frequent impairment, the plains reference sites probably represent the potential of this region for supporting aquatic uses. In the mountain region, channel condition and use support may vary considerably between the upper and lower reaches of a stream. Streams in the foothill region have a good potential to fully support aquatic uses through improved land management practices. In several streams, the sample sites were not located on the reach that was assessed as least impaired.

INTRODUCTION

This report describes the composition and structure of benthic macroinvertebrate (mostly insect) and periphyton (algae) communities inhabiting least-impaired reference streams in Montana's six major ecoregions in the summer of 1990. The report also describes the fish fauna of these streams and provides supporting information on water chemsitry, macroinvertebrate habitat, and overall stream condition.

BACKGROUND

There is little of this earth that has escaped the hand of man. From polar seas to tropical forests, the biosphere has been tainted chemically, altered physically, and diminished in biological diversity (Gore 1992). With these changes have come diminished natural beauty, a loss of species and productivity, and, in some cases, a collapse of ecosystem function.

Even in Montana, among the least populated and industrialized of the United States, all of the larger lakes and rivers have suffered some degree of use impairment (DHES 1990). Most of this impairment is caused by a pervasive and multifaceted genre of pollution called "nonpoint." Agriculture, forest practices, mining, dams, dewatering, habitat alterations, and land disposal account for about 90 percent of the impaired waterbodies in Montana. Point source discharges from municipal and industrial wastewater treatment plants account for the remaining 10 percent.

The goal of the 1972 federal Clean Water Act is to restore and maintain the chemical, physical, and biological integrity of the nation's waters (Section 101). The Act further instructs the states and the U.S. Environmental Protection Agency (EPA) to maintain a level of water quality that will assure protection and propagation of balanced indigenous populations of fish and aquatic life (Section 303). More recently, EPA's Science Advisory Board (1990) recommended that EPA give as much attention to addressing ecological risk as it does to human health risk because "over the long term, ecological degradation....degrades human health and the economy."

RATIONALE

Until now, water pollution control programs have been based primarily on the measurement of chemical constituents and, to a lesser extent, on laboratory tests of the toxicity of wastewater discharges to selected organisms. While these approaches did help to reduce concentrations of many water quality contaminants, they did not directly gauge the health of whole biological communities in aquatic ecosystems, most notably those subject to the cumulative effects of both point and nonpoint sources of pollution.

Recognizing this need, EPA has begun to emphasize a more ecological approach to water pollution control (Figure 1). Among the agency's recent initiatives have been the development of ecoregions (Omernik 1986, 1987), protocols for rapid bioassessment (Plafkin et al. 1989), guidelines for developing biological water quality criteria (EPA 1990), and a nationwide surface water ecology monitoring and assessment program (Paulsen et al. 1991). States have been instructed to develop narrative biological criteria by 1993; numerical biological criteria will come later.

Plafkin et al. (1989) describe several advantages of sampling biological communities in water quality investigations:

- 1. Biological communities reflect overall ecological integrity. Ecological integrity is defined as the "ability to support and maintain a balanced, intergrated, adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of natural habitat of the region." (Karr and Dudley 1981).
- 2. Biological communities integrate the effects of different pollutant stressors and thus provide a holistic measure of their aggregate impact over time.

- 3. Routine monitoring of biological communities can be relatively inexpensive, particularly when compared to the cost of assessing toxic pollutants using chemical analyses or toxicity test (bioassays).
- 4. The status of biological communities is of direct interest to the public as a measure of a pollution-free environment.
- 5. Where criteria for specific ambient impacts do not exist (e.g., nonpoint-source impacts that degrade habitat), biological communities may be the only practical means of evaluation.

PURPOSE

The Montana Water Quality Bureau has three reasons for establishing benchmark or baseline biological conditions in least-impaired reference streams:

- To provide a reference against which to compare conditions in other streams. Such a
 reference will help gauge the severity of pollution as well as progress in abating
 pollution. This will be particularly helpful in the Nonpoint Source Program- for ranking
 prospective watershed demonstration project streams and measuring the effectiveness of
 best management practices.
- 2. To provide the basis for narrative and numerical biological criteria and enforceable biological standards in streams.
- 3. To describe the natural biodiversity and types of algal and macroinvertebrate communities found in Montana streams. The concept of biodiversity recognizes the intrinsic value of biological species and their functional importance in ecosystems. Little work of this type has been accomplished in aquatic ecosystems as compared to terrestrial ecosystems; and few studies of biodiversity have addressed invertebrates and nonvascular plants as compared to vertebrate animals and vascular plants.

This report will be followed by three others: (1) A report based on sampling in 1991 describing year-to-year, reach-to-reach, and riffle-to-riffle variation in biological metrics in selected Montana reference streams; (2) A report recommending narrative and numerical biological criteria for Montana streams; and (3) A report recommending protocols for assessing water quality conditions in Montana streams using biological metrics.

SCOPE AND LIMITATIONS

Limited resources restricted the scope of this study in several respects. As such, the results presented here can be considered only a first approximation and not the definitive account of the range of biological characteristics found in Montana's least-impaired streams.

- This report addresses only second to sixth order streams as defined by Strahler (1957). It
 does **not** address smaller streams (first order streams or spring seeps), larger streams or
 rivers, lakes, ponds, reservoirs, springs, or wetlands. Larger streams and rivers in
 Montana are generally impaired (DHES 1990) and the biological characteristics of the
 other types of waterbodies mentioned above are sufficiently different to warrant separate
 investigations.
- 2. The streams selected may not represent the entire range of stream gradient for streams within each ecoregion.
- 3. The findings reflect conditions only in summer (June 21 to September 21). Summer was chosen because (1) periphyton diversity in Montana streams peaks in summer and early fall (Bahls et al. 1979a, 1979b, 1981; Ingman et al. 1979), (2) Montana streams generally

have stable flows in summer, and (3) summer is the most temperate and least troublesome time for aquatic field work in Montana.

- 4. This report addresses only the structure and composition of the periphyton and macroinvertebrate communities. It does not address the biomass, chemical quality or functional aspects of these communities, which include such features as productivity, standing crop, toxic residues, respiration, and photosynthesis. And beyond a listing of indigenous and introduced fish species present, it does not address other biological groups, such as macrophytes and protozoans.
- 5. This report addresses biological conditions at only a single riffle in a single reach of each stream during one year (1990). A follow-up report, based on samples collected in 1991, will describe year-to-year, reach-to-reach, and riffle-to-riffle variations in selected streams.

STUDY DESIGN

MONTANA ECOREGIONS AND SUBREGIONS

The ecoregions of Omernik and Gallant (1987) were chosen as the basis for spatially stratifying reference streams to account for geographical variation in Montana. These ecoregions are based on patterns of land use, land-surface form, potential natural vegetation, and soils.

Portions of seven ecoregions occupy the State of Montana (Map 1). Six of these occupy large areas of the state:

- Northern Rockies (#15)
- Montana Valley and Foothill Prairies (#16)
- Middle Rockies (#17)
- Northern Montana Glaciated Plains (#41)
- Northwestern Glaciated Plains (#42)
- Northwestern Great Plains (#43)

A small portion of the Wyoming Basin (#18) is located in southcentral Montana.

The Northern Rockies and Middle Rockies were divided into subregions to address the effects of significantly different geologic formations (e.g., granitic and non-granitic rocks) and opposing slopes of the Continental Divide. Spring Creeks were singled out as a subset of streams in the Montana Valley and Foothill Prairies. The remaining streams in this ecoregion, the "special creeks", were selected for their potential to serve as reference streams for existing and planned Section 319 nonpoint source watershed demonstration projects, most of which are also located in this ecoregion. The streams in this special group-Elk Creek, Big Hole River, and Big Spring Creek- later proved to be threatened or impaired and less than ideal for this purpose.

STREAM SELECTION

Our goal was to sample 35 to 40 streams during the summer of 1990, or about 6 or 7 streams in each ecoregion. The first step in selecting these streams was to compile a list of candidate streams. This was done by mailing a nomination form and instructions to Montana Department of Fish, Wildlife and Parks (DFWP) Regional Fisheries Biologists, Conservation District Supervisors, Forest Service Hydrologists, Soil Conservation Service (SCS) Area and District Conservationists, and Bureau of Land Management (BLM) District Managers. These people were asked to nominate two least-disturbed streams in each of three size classes: small stream (stream order 2); mid-size stream (stream orders 3 and 4); and large stream/small river (stream orders 5 and 6). They were advised that the streams' watershed and riparian zones should be minimally disturbed compared to other streams in the area, but that the watershed need not be in pristine condition. Assured long-term protection of the stream's watershed was not a criterion of selection. Further, the streams should have flow in summer and road access.

Fifty-four nomination forms were completed and returned. To the list of nominated streams were added streams known to the monitoring staff of the Water Quality Bureau. The streams were then sorted by ecoregion and subregion. The final selection was based on the following criteria:

- 1. Information on fish species exists in the DFWP Interagency Stream Fisheries Database.
- 2. Stream is not water quality limited, i.e., sample reach does not appear on the list of impaired streams in the 1990 Montana 305(b) Report.

- 3. Stream lies within the "most typical" area of each ecoregion as defined by Omernik and Gallant (1987).
- 4. One small stream (order 2), one mid-size stream (order 3 or 4), and one large stream (order 5 or 6) should be sampled in each ecoregion or subregion. (While smaller streams in the mountain ecoregions tend to have steeper gradients, a conscious effort was not made to select streams representing the range of gradients within an ecoregion.)

The 38 streams selected for sampling (Table 1 and Map 2) represent some of the last best streams in Montana. There are many more equally good reference streams in the mountain ecoregions. On the other hand, it was quickly apparent that not all of the selection criteria could be satisfied for most streams in the plains ecoregions. Two streams in the Wyoming Basin- Jack Creek and Cottonwood Creek- were visited but not sampled; neither was flowing and both were void of macroinvertebrates at the sampling site. (Jack Creek was later assessed as severely impaired and Cottonwood Creek as not impaired but threatened.) Some streams (e.g., Elk Creek, Pine Creek, Willow Creek) were later excluded from some evaluations of ecoregional characteristics when field assessments and examination of the data revealed various degrees of impairment in these streams.

SITE SELECTION

Sampling sites were designated on county or topographic maps prior to field work. A primary consideration was accessibility by road. An effort was made to locate sites upstream from impoundments and areas of human disturbance. In western Montana, sites were generally located at the boundaries of roadless areas or National Forests. Some sites had to be changed when field reconnaissance revealed access problems (closed roads) or previously undetected perturbations. Stream locations are shown on Map 2 and sampling site locations are described in Table 1.

BIOLOGICAL GROUPS

Several functional or taxonomic groups ("communities") or organisms exist in a typical Montana stream. These include decomposers (bacteria and fungi), producers (macrophyton and periphyton), and consumers, including microinvertebrates (invertebrates too small to be seen by the unaided eye), macroinvertebrates (invertebrates that can be seen by the unaided eye), and fish.

Periphyton (benthic algae) and macroinvertebrates were selected for sampling and detailed water analysis in this survey for several reasons: (1) the Water Quality Bureau has inhouse expertise and experience in using both groups; (2) they are practically ubiquitous in Montana streams and represented in unpolluted streams by relatively large numbers of species and individuals; (3) standard methods of analysis and a large body of taxonomic and ecological literature exist for both groups; (4) sampling is relatively quick and inexpensive and creates minimal impact on the stream's habitat and resident biota; and (5) they span several trophic levels and are sensitive to a wide range of environmental pollutants. Plafkin et al. (1989) list additional advantages to using benthic macroinvertebrates and algae in biosurveys.

There are also several advantages to using fish (Plafkin et al. 1989). Available information on fish populations is included in this report recognizing the importance of fish in stream ecology, in environmental law ("fishable and swimmable"), and to the fishing public. This information is used primarily for stream classification (e.g., warm water and cold water fisheries) and to describe in general terms the species composition and species richness of fish communities in each of the ecoregions. The Water Quality Bureau is neither equipped nor authorized to collect fish in Montana, and resources were not sufficient to contract for the sampling and evaluation of reference stream fish communities.

BIOLOGICAL METRICS

Much can be learned about a community of aquatic organisms and the quality of its aqueous environment simply by reviewing a list of the species present. Certain sensitive species indicate good

water quality and some tolerant species may indicate poor water quality, especially if they are represented by a large number of individuals. However, review of species lists is a tedious and subjective process and it does not address important structural features of the community. Biological metrics are a quick and convenient way to summarize and merge taxonomic and structural features.

A biological metric or indicator is a shorthand numerical representation of a biological community. To be useful for water quality monitoring, a metric should change in a predictable manner to changing water quality conditions. Perhaps the most universally used metric is "species richness": the number of species in a sample of organisms of a given taxonomic group. As a rule, species richness is directly correlated with water quality as water quality declines, so does the number of individuals or the number of species that are either tolerant or intolerant of certain kinds of pollution.

Metrics may be positively or negatively related to water and habitat quality. For example, EPT Richness -- the number of species in a largely pollution sensitive macroinvertebrate orders Ephemeroptera, Plecoptera and Trichoptera -- is positively related because values increase as water and habitat quality increase. On the other hand, the Hilsenhoff Biotic Index is negatively related because values increase as water and habitat quality decrease.

For macroinvertebrates, a wide variety of metrics were calculated, including metrics from Plafkin et al. (1989) and Barbour et al. (1992). Standard periphyton metrics include diatom species richness and Shannon diversity (Bahls 1979), and percent relative abundance (PRA) of diatom species in different pollution-tolerance groups (Lange-Bertalot 1979). From an examination of Lange-Bertalot's initial listing of taxa, these groups are evidently based on tolerance of or sensitivity to several classes of pollutants, including temperature, nutrients, sediments, toxics, and salts. Diatom species not listed by Lang-Bertalot were assigned to pollution tolerance groups on the basis of the autecological literature (e.g., Lowe 1974 and Bahls et al. 1984) and data collected previously by the Water Quality Bureau.

Additional periphyton metrics are proposed and calculated in this report on an experimental basis. A rationale for these metrics is given here:

- Number of genera of common non-diatom algae (genus richness): Same rationale as for number of diatom species -- more taxa mean better community stability and higher quality water.
- 2. Percent relative abundance (PRA) of the dominant diatom species: Communities in polluted waters are generally dominated by one or a few species having a large number of individuals. This metric is equivalent of Metric 5 in Plafkin et al. (1989).
- 3. PRA and number of species of *Navicula* and *Nitzschia*: among the genera of diatoms observe to have motile species (Werner 1977), *Navicula* and *Nitzschia* are by far the most common in Montana streams, both in number of species and number of individuals. Motile species are adapted to maintaining their position on unstable substrates, so the proportion of motile species in a sample may be directly related to the degree of sedimentation on the stream bottom at that site.

SUPPORTING INFORMATION

Biological surveys are incomplete without supporting information on water quality, habitat, and overall stream condition. This information is needed to understand the factors that regulate biological communities and determine the value of biological metrics.

Three types of supporting information were gathered:

- A suite of chemical and physical water quality variables. Temperature, pH, and dissolved oxygen were measured in the field while streamflow was estimated visually. Variables measured in the laboratory included common ions, nutrients, specific conductance, total hardness, total suspended sediment, and pH.
- 2. As assessment of physical habitat at the sampling site, adapted from Plafkin et al. (1989).
- 3. As assessment of overall stream conditions (generally of the reach containing the sampling site) using the Water Quality Bureau's Nonpoint Source Stream Reach Assessment technique (Appendix 1) and Numerical Ranking Criteria (Appendix 2).

This information will be used to help classify streams and to explain variation in biological metrics.

METHODS

WATER QUALITY

Field

Water samples for laboratory analysis were "grabbed" by dipping a polyethylene bottle in the main current of the stream to mid-depth. All sample collection, handling, preservation, and storage procedures followed EPA-approved methods described by DHES (1989).

Streamflow rates were estimated visually. Water temperatures were measured with calibrated mercury field thermometers having a resolution of 0.1 C. Field measurements of pH were made with a portable meter accurate to 0.01 standard unit. Dissolved oxygen was measured by the Winkler method.

Laboratory

All laboratory analyses were performed by the Chemistry Laboratory Bureau of the Montana Department of Health and Environmental Sciences. All variables were analyzed by EPA methods for Chemical Analysis of Water and Wastes (1983).

STREAM REACH ASSESSMENTS

Field Assessment

Assessments were completed by three individuals representing OEA Research of Helena, Montana using protocols set forth in the NPS Stream Reach Assessment Field Form developed by DHES/WQB (Appendix 1). The form is designed to allow assessment to both natural (including hydrologic) and human influences on stream conditions.

The assessment form contains 22 indicators of stream conditions, ranging from general (e.g., land use) to specific (e.g., water odor). The form also contains one page for describing and evaluating the implementation of best management practices (BMPs), which are activities designed to protect water resources from nonpoint source pollution. This section provided supporting information for assessing individual indicators, but is not used for assessing overall impairment.

The field assessment procedure entails spot check observations at selected points within each stream reach. To save time, the assessors usually chose observation points that were accessible by road or within a short walk from a road. Assessments of unroaded reaches were often based on limited observation points. An experienced assessor can assess 10 to 20 miles of stream per day, depending on accessibility.

Relatively homogenous stream reaches were defined by the assessors and based on a combination of factors, such as stream gradient, valley bottom shape, vegetation, substrate, stream order, and land use. Prior to conducting field work, the assessors used aerial photos (when available) and topographical maps to identify possible stream reaches and observation sites. Final determination of reaches and observation points was made in the field.

At each observation point, the assessor attempted to observe at least one riffle, one run, and one pool. Adjacent land features, and land management practices were also assessed. The condition of each indicator listed on the assessment form was noted at each observation point. An assessment form was completed for each reach. The assessment was based on information compiled from all observation points within that reach.

Overall Impairment and Use Support

For the purpose of this assessment procedure, impairment is defined as the degree to which a stream will support beneficial uses. Beneficial uses are those designated in the Water-Use Classifications of the Montana Surface Water Quality Standards (ARM 16.20.604 through 16.20.612).

Numerical criteria were developed to assess the overall level of impairment and use support within each reach (Appendix 2). Each indicator of stream condition- bank stability for example- is divided into three or four subjective descriptions ranging from poor to excellent. Each description is assigned a number from 1 to 4 with larger numbers representing better conditions.

To determine overall impairment and use support for each reach, the sum of values for all of the assessed indicators is divided by the total points possible for those indicators. This value is multiplied by 100, yielding a percentage. The indicators that are actually assessed and the total points possible may vary from reach to reach depending on conditions encountered during the assessment (for example, extreme turbidity may preclude substrate assessment). The overall assessment it then converted into one of five impairment and use support categories as follows:

87 - 100% = Not impaired (full support)

80 - 86% = Not impaired, but threatened (full support)

71 - 79% = Minor impairment (partial support)

56 - 70% = Moderate impairment (partial support)

0 - 55% = Severe impairment (non-support)

These assessments are intended to provide a rapid and general overview of stream conditions and factors influencing those conditions. They can be used to estimate stream impairment and use support when more intensive surveys are not affordable. Actual levels of impairment and use support are more accurately determined by methods such as water quality monitoring, biomonitoring, bioassessment, and bioassays.

PERIPHYTON

The following methods for collecting and analyzing periphyton have been used by biologists at the Montana Water Quality Bureau since 1973, for surveillance monitoring, intensive surveys, and fixed-station monitoring. These methods generally involve about 3 hours total elapsed time per sample and do not require a return trip for retrieving artificial substrates. The two-step analysis procedure provides (1) a ranking of non-diatom genera by relative volume (biomass) and (2) the percent relative abundance of individual diatom species. No effort is made to ascertain the proportion of dead (empty) diatom frustules because these are considered to be part of the "history" of the stream periphyton community from the time stream-bottom substrates were last scoured and pioneered with algae.

<u>Field</u>

Periphyton was collected following Procedure 6.2.2 in the Field Procedures Manual of the Montana Water Quality Bureau (DHES 1989). This entails collecting microalgae (algae living as single cells or in microscopic colonies) from **natural** substrates (rocks, logs, moss, mud) in proportion to the area covered by those substrates at a given access site. This commonly entails scraping the entire surface of two or more rocks of different sizes selected at random. Portions of macroalgae (algae growing in large filaments or colonies visible to the naked eye) are collected in proportion to their abundance at that site. While effort is usually concentrated in riffles, other macrohabitats (pools and runs) are also sampled if they support algae growth. The goal is to collect a single composite sample that is a miniature of the stand of algae that is present at that site.

Samples were placed in small, screw-top jars and preserved with enough iodine potassium iodide (Lugol's solution) to impart a reddish-brown tint. They were then transported to the laboratory and stored in a refrigerator until the time of the analysis.

Laboratory

Analysis of the samples was carried out in two stages: the first for soft-bodied (non-diatom) algae and the second for diatoms. Analyses were performed by Erich Weber of PhycoLogic in East Helena, Montana.

Each sample was poured into a white enameled pan and portions of conspicuous macroalgae were removed to a microscope slide having a shallow central basin. The remainder of the sample was returned to the sample jar and thoroughly agitated (to randomize algae cells and colonies) and then, using a soda straw, a several-drop subsample of microalgae was added to the fragments of macroalgae in the shallow basin. A coverslip was placed over the shallow basin, completing a composite wet mount.

Each wet mount was scanned under a WILD standard research microscope, first at 200X and then at 400X. Soft-bodied algae were identified to genus using primarily Smith (1950) and Prescott (1978). After all of the common soft-bodied algae were identified, each genus was ranked according to its estimated contribution to the total volume or biomass of the sample, taking into account the remaining macroalgae and microalgae in the original sample. Diatoms were included, but they were ranked as a group (Class Bacillariophyceae) and not as individual genera. Genera that were rated rare (see below) were not ranked.

Genera of soft-bodied algae and diatoms as a group were also rated as to the relative abundance of their cells:

- R (rare) fewer than one cell per microscope field at 200X, on the average;
- C (common) at least one, but fewer than five cells per field of view;
- VC (very common) between 5 and 25 cells per field;
- A (abundant) greater than 25 cells per field, but numbers within limits reasonably counted
- VA (very abundant) number of cells per field too numerous to count

These designations have no counterpart in terms of cells per unit area of stream bottom. Although the density of algal material in each wet mount will vary, a certain degree of standardization is achieved by the need to provide sufficient separation of cells and passage of light through the mount to allow for identification of genera and estimation of cell numbers.

The next step was to prepare the remainder of the sample for diatom analysis. The sample was thoroughly agitated once again (to dislodge diatom epiphytes) and about 25ml of sample was poured immediately into a 100ml glass beaker. Under a fume hood as small amount of concentrated sulfuric acid and then potassium dichromate were added to each beaker to oxidize the organic content of the sample, leaving behind inorganic sediment and "cleaned" diatom frustules. (**Caution!** This will usually cause a strong exothermic reaction.) Acid and dichromate are added until they no longer cause any visible reaction. This procedure effectively breaks up diatom colonies and randomizes diatom cells.

The beakers were left to cool, then filled with distilled water and allowed to stand overnight. The supernatant was then poured off and each beaker was again filled with distilled water and this time left to stand a minimum of 4 hours. This procedure was repeated until all trace of color was gone from the supernatant. Using the "cleaned" residue, a permanent diatom strewn mount was prepared as described in "Standard Methods" (APHA et al. 1980).

Each diatom slide was scanned under 1000X (100X oil immersion objective, 1.30 numerical aperture) and a list of species was compiled using primarily Patrick and Reimer (1966, 1975), Krammer and Lange-Bertalot (1986, 1988) and Hustedt (1930) for identification. A diatom proportional count was then performed according to "Standard Methods" (APHA et al. 1980), generally counting between 350 and 400 diatom cells (frustules). Counts for each species were divided by the total count and multiplied by 100 to obtain percent relative abundance (PRA). Those species encountered in the floristic scan but not during the proportional count were designated with a "p" for present.

MACROINVERTEBRATES

Field

The macroinvertebrate sampling unit was a representative cross section of the microhabitats available within a riffle. In most cases, the sampler employed a diagonal travelling kick technique using a D-frame aquatic net with 1mm mesh. In streams that had neither current nor riffles it was necessary to use the net like a sweep net. The samples were meant to be qualitative, although the sampling effort was timed and the length of stream bottom disturbed was estimated to allow for estimating "catch per unit effort". Samples were preserved in the field with 95% ethanol to approximate a final sample concentration of 80% ethanol.

Laboratory

Macroinvertebrate samples were processed and data analysis conducted by Aquatic Biology Associates of Corvallis, Oregon. The samples were processed as follows:

- 1. Samples were cleaned of fine sediment by gently washing on a 500-micron sieve.
- 2. The entire sample was then floated in water in a shallow white pan and large debris rinsed and removed.
- 3. Organic matter and invertebrates in the pan were repeatedly suspended in water and poured off on to a 500-micron sieve. The mineral material remaining in the pan was searched for organisms that did not float off during the washings, such as snails and stone-cased caddis larvae.
- 4. Large samples were split into half or quarter fractions using a Jones Riffle Splitter. A fraction of containing about 300 organisms was randomly selected for further sorting. The appropriate coefficient was later used to adjust taxa abundances to a full kick sample.
- 5. Invertebrates were sorted from a sample matrix using a dissecting microscope at 6X magnification.

Standardization of Taxonomic Effort

Invertebrates were identified, counted, and recorded on bench sheets. Then a "master list" of taxa was produced from the completed bench sheets. This "master list" was combined with lists from other Montana macroinvertebrate studies to produce a Montana Macroinvertebrate Checklist (Appendix 3).

For each grouping of taxa, a decision was made as to the appropriate level of taxonomic resolution. Emphasis was placed on consistency of identification. In most cases identifications were made at a level that would standardize taxonomic effort between samples. The level of taxonomic resolution employed for the macroinvertebrate portion of this study is outlined in Appendix 4.

A data entry sheet was made listing the standardized taxa, and data from the original bench sheets were transcribed and adjusted accordingly. For some sites, finer levels of taxonomic resolution were still

used in data analysis. In these cases only a single taxon was involved, so the difference would not influence the computation of metrics. For example, *Gyraulus* was used instead of *Planorbidae*.

Data Analysis

Spatial patterns exhibited by macroinvertebrate communities were explored with an ordination technique called Detrended Correspondance Analysis or DECORANA (Gauch 1982, Hill 1979).

DECORANA is a computer program designed to summarize the information contained in complex sets of data from biological communities. The information produced is summarized by locating each site on a two-dimensional plot. DECORANA groups the sites according to similarity such that similar sites are near each other and dissimilar sites are far apart.

DECORANA was run on a matrix containing percent relative abundance data for all taxa from all riffle macroinvertebrate samples. Data from a relatively health site on Big Otter Creek (Montana Valley and Foothill Prairies Ecoregion) were included in the analysis.

Results of the DECORANA were used to select sites for constructing regional reference communities. These composite communities were used to calculate the similarity index values presented in this report and to summarize the dominant regional taxa. They will likely be modified as more data become available.

The composite regional reference communities were derived as follows:

- 1. For each region, a matrix of percent relative abundance values was constructed for all taxa and all sites. Average values were used for streams in which two sites were sampled. Big Spring Creek was excluded from the analysis because it did not group with any other stream.
- 2. Average percent relative abundance values for each taxon were then derived for the entire regional data set.
- 3. Taxa were ranked by average abundance, from most abundant in the region to least abundant.
- 4. The resulting composite lists of taxa varied from 100 to over 200 taxa per region. In order to obtain more realistic comparisons for calculating similarity indices between specific sites and the composite reference community, the composite community was limited to the number of most dominant taxa equal to the mean number of taxa found in streams in that region.

Computer programs written in Fortran analyzed the raw data to compute the metrics. The composite reference stream communities were compared to the raw data for each site in the region to calculate the various similarity indexes. About 35 metrics were calculated for each site sampled. These include the ones proposed by Plafkin et al. (1989), Shackleford (1988) and others.

Not all of these 35 metrics need to be or should be used for developing biocriteria or for conducting routine bioassessments, so a subset of metrics was selected for treatment in this report. Lacking a database from Montana or Northwest streams for evaluating the response and sensitivity of individual metrics to a variety and range of impairments, the following criteria were considered in selecting this subset:

- 1. Has the metric had historical use as a water quality indicator?
- 2. Is the variability of the metric relatively low among reference of least-impaired streams in a region?

- 3. Is the metric applicable in a large number of the reference streams in a region? (Some metrics calculated from ratios have a large number of zero or undefined values in some regions).
- 4. Do the selected metrics include the several classes of community descriptors, including taxa richness, taxonomic composition, functional feeding groups, diversity, dominance, and pollution indicators?

Macroinvertebrate Habitat

A modified habitat assessment as outlined in Plafkin et al. (1989) was conducted at each site (Appendix 5). Total habitat assessment scores were plotted against selected macroinvertebrate community metric values to check for correlations.

FISH

Information on fish species present, their relative abundance, and their use of the reference streams, was obtained from the Interagency Stream Fishery Database maintained by the Department of Fish, Wildlife and Parks, and from regional fish biologist employed by this agency. Fish species were designated either native or introduced according to information contained in Brown (1971).

RESULTS AND DISCUSSION

WATER QUALITY

Results of all water quality analyses are presented in Appendix 6. Results of selected key variable are presented and discussed below.

Major Ions

Calcium was the major cation and bicarbonate the major anion in streams of the mountain ecoregions, including those of the Montana Valley and Foothill Prairies (Table 2). In streams of the plains ecoregions, sodium was the major cation and either sulfate or bicarbonate was the major anion.

Physical Variables

Except for the West Gallatin River, estimated flow rates of Montana reference streams were all less than 150 CFS (Figure 2). Flow rates of plains streams were signicantly smaller than flow rates of mountain streams.

Except for Elk Creek, water temperatures in the mountain streams were all below the Montana surface water quality standard of 19.4 C for protecting cold water aquatic life (Figure 3). Water temperatures in the plains streams were all below the standard of 26.6 C for protecting warm water aquatic life.

Specific conductance was less than 500 umhos/cm in all mountain reference streams and greater than 500 umhos/cm in all plains reference streams (Figure 4). Based on a freshwater threshold of 1500 umhos/cm, all mountain reference streams were freshwater and plains reference streams were either freshwater or brackish.

Concentrations of suspended sediment tended to be much larger and more variable in plains reference streams than in mountain reference streams (Figure 5). All mountain reference streams had sufficiently low concentrations of suspended sediment to afford a "high level of protection" to fish and aquatic life based on criteria published by EPA (1973). Protection in plains reference streams ranged from "high" to "low".

PH, Alkalinity, Hardness

All Montana reference streams had pH values larger than 7.00 (Figure 6). Streams in the West Side/Granitics and Absaroka-Beartooth subregions had pH values ranging from 7.00 to 8.0; other streams generally had pH values in the range of 8.00 to 9.00 standard units.

Alkalinity- a measure of a water's ability to neutralize acid- was higher in the plains reference streams (Figure 7). Among mountain reference streams, those in the West Side/Granitics and Absaroka-Beartooth subregions would be the most sensitive to acid mine drainage or acid deposition due to generally low alkalinity values.

Total hardness- a measure of a water's ability to precipitate soap and neutralize the toxicity of heavy metals- was also higher in the plains reference streams (Figure 8). Reference streams in the Granitics, West Side/Non-Granitics, and Absaroka-Beartooth subregions had generally "soft" waters that would be very sensitive to contamination by heavy metals. Reference streams in the plains ecoregions generally had "hard" to "very hard" waters that would be relatively insensitive to the effects of heavy metals.

Nutrients

Mean total phosphorus and nitrogen values were higher in the Montana Valley and Foothill Prairies and the plains ecoregions (Figures 9 and 10). The Northern Montana Glaciated Plains Ecoregion had by far the largest maximum and average values for total P and N.

Concentrations of bioavailable P and N (ortho-P and total inorganic N, respectively) in Montana reference streams are compared in Figures 11 and 12 with values known to allow standing crops of nuisance algae in the Clark Fork River (Watson 1990). Mean values tend to fall below the nuisance thresholds in all reference streams except those in the Montana Valley and Foothill Prairies (P and N) and in the plains ecoregions (P only).

The ratio of total inorganic nitrogen to ortho-phosphate as P is plotted in Figure 13. The solid vertical line represents the optimum ratio of bioavailable N to bioavailable P for plant growth: 10 to 1.

Streams to the right of this line- mostly mountain streams- tend to be low in phosphorus relative to the amount of nitrogen available for plant growth, although both nitrogen and phosphorus are in short supply compared to plain streams. These streams would respond most dramatically to an increase in phosphorus or a combination of phosphorous and nitrogen.

Streams to the left of the solid vertical line- mostly plains streams- tend to be poor in nitrogen relative to the amount of phosphorus available for plant growth. These streams would respond most dramatically to an increase in nitrogen.

Dissolved Oxygen

Dissolved oxygen concentrations exceeded Montana Surface Water Quality Standards in all of the reference streams for this variable (Appendix 6).

STREAM REACH ASSESSMENTS

Although the entire reference stream was assessed in most cases, results are reported here only for the reach that includes the sample site. Thus "stream" in this section refers to the reach that includes the sample site rather than to the entire reference stream. Results of the assessments are summarized in Appendix 7.

Use impairment and support status for the 38 assessed reference streams were as follows:

10 streams (26%)	Not impaired (fully supporting)
8 streams (21%)	Not impaired, but threatened (fully supporting)
12 streams (32%)	Minor impairment (partially supporting)
7 streams (18%)	Moderate impairment (partially supporting)
1 stream (3%)	Severe impairment (non-supported)

All ecoregions contained one or more partially supporting (impaired) reference streams. However, all streams in several subregions were assessed as fully supporting (not impaired). These subregions included the Granitics and West Side/Non-Granitics subregions of the Northern Rockies Ecoregion, and the Absaroka-Beartooth subregion of the Middle Rockies Ecoregion. Assessed impairment and use support ratings for each reference stream are presented in Table 3.

Ecoregion Summaries

The following discussion focuses on impaired (partially or non-supporting) reference streams. Streams not mentioned by name were assessed as fully supporting. Appendix 7 contains complete assessment summaries for all reference reaches, by ecoregion.

Northern Rockies. Of the nine streams assessed in the Northern Rockies ecoregion, only Waldron Creek and the North Fork of the Teton River did not fully support beneficial uses, primarily due to bank and channel instability. Land use is limited within these reaches (both emerge from the Bob Marshall Wilderness) and instability problems are apparently related to 1964 flood damage along the Rocky Mountain Front.

Contrary to what might be expected, all reference streams in the highly erosive Granitics subregion were unimpaired and fully supporting.

<u>Middle Rockies.</u> The West Gallatin River, Pine, Tenderfoot and Calf Creeks were assessed as partially supporting their uses. Grazing occurs on Tenderfoot and Calf Creeks, causing bank instability and damage to riparian vegetation. Pine Creek suffers from heavy sedimentation and bank and channel instability. Sedimentation is due in part to bank erosion caused by instream debris jams. Road erosion and upstream logging may also contribute sediment. The West Gallatin is affected primarily by highway encroachment and localized subdivision activity.

Montana Valley and Foothill Prairies. Ben Hart Spring Creek, Odell Spring Creek, Elk Creek and Big Spring Creek were assessed as partially supporting their uses. Agricultural activities, primarily grazing, occurred along each of these streams. Grazing effects on bank stability and streamside vegetation were the primary causes of stream impairment.

Northern Montana Glaciated Plains. All three streams in this ecoregion were partially supporting. Rock Creek was assessed as having minor impairment; Battle and Willow Creeks were moderately impaired.

This region is characterized by an erosive, poorly vegetated landscape. For all three streams, impairment was attributed primarily to natural soil erosion and bank instability. Natural erosion was aggravated by grazing. When the assessments were conducted in October and November, all three streams were significantly dewatered and water was confined mainly to isolated pools or downstream reaches. Moderate salinization was evident in Rock and Willow Creeks.

Northwestern Glaciated Plains. Tule Creek and the Redwater River were assessed as partially supporting with both having minor impairment. Common factors contributing to the impairment of these two streams were dewatering, lack of stream shading, and bank instability due primarily to grazing. Naturally erosive soils also contributed to bank instability. Natural erosion in the "breaks" area of the Redwater River was greater than natural erosion along Tule Creek. Moderate salinization was also observed in the Redwater.

<u>Northwestern Great Plains.</u> Four of the five streams in this ecoregion were assessed as partially supporting. Hanging Woman and O'Fallon Creeks exhibited minor impairment; Larb Creek and the Tongue River were moderately impaired.

Impairment in this ecoregion was generally related to moderate levels of natural upland and streambank erosion aggravated by grazing and minor amounts of cultivation. Grazing and cultivation also impacted streamside vegetation. The extent of natural erosion was the primary influence separating degree of impairment. Salinization was moderate to severe in Larb, Hanging Woman and O'Fallon Creeks. Dewatering was pronounced in all streams except Tongue River.

Wyoming Basin. Both reference streams in this ecoregion are impaired. Cottonwood Creek was assessed as partially supporting uses due primarily to cultivation encroaching on streamside vegetation. Upstream salinity and grazing may also influence conditions at the reference site.

Jack Creek was assessed as non-supporting (severely impaired). This stream was dry at the time of the assessment and grass and sagebrush in the channel indicated the stream rarely flows. Standing water was observed in culvert scour holes at the road crossing located three miles above the mouth. Grazing had impacted riparian vegetation and bank stability. A white precipitate was observed, indicating salinization.

Ecoregion Groups

Due to similarities in stream features, results have been grouped into three major regions: mountain, foothills, and plains. These three regions represent the three major geographical areas or physiographic provinces of the Montana landscape.

The Montana Valley and Foothill Prairies Ecoregion equates to the foothills region. This includes the spring creeks and "special creeks". The mountain region contains the Northern and Middle Rockies Ecoregions. The Wyoming Basin and the three plains ecoregions comprise the plains region. To evaluate potential use support for each stream, it was necessary to analyze assessment results for the entire stream rather than for just the reference reach. In contrast to the preceding discussion, the term "stream" in this section refers to the entire reference stream rather than to the reach containing the reference site.

<u>Mountain Region</u>. The majority of streams in the mountain region are unimpaired and likely represent this region's potential relative to support of aquatic life. The exceptions in the Middle Rockies Ecoregion all had management activities occurring within the reference reach. These streams are probably not at their potential and alternative reference sites should be considered.

Two streams in the East Side subregion of the Northern Rockies Ecoregion-Waldron Creek and the North Fork of the Teton River- are impaired due to instability apparently caused by the 1964 flood. Perhaps better (i.e., more stable) reference streams are not available in this subregion because of this major flood.

Compared to the other regions, stream variability is more pronounced in the mountain region. Stream character may change dramatically between the steep headwaters and flatter upper valley bottoms. The assessments generally revealed that the upper stream reaches were slightly less impaired than the lower reaches. This was due mainly to more concentrated land use activities in the lower reaches. However, channel type also may have influenced the impairment values. The lower reaches generally exhibited lower gradient, greater channel sinuosity, and a higher incidence of bank and channel instability. These lower reaches may exhibit characteristics more similar to streams in the foothills region.

Foothills Region. Natural conditions in the foothills region do not appear to significantly influence impairment values. These streams have good potential to support healthy and diverse aquatic communities. The impacts to aquatic uses exhibited in this region are closely tie to management activities. Controlling impacts from management activities is feasible, as demonstrated on lower Odell Creek (reference: P. Hackley, stream reach assessment notes). Management activities occurred on all of the streams in this ecoregion, but some streams were less impaired than others. These differences may be due to the level or type of management, or to different vegetation and soil types that temper or enhance management impacts.

Less impaired reference streams may be difficult to find in this region due to the unique nature and limited availability of spring creeks and the prevalence of agriculture. In some cases, less impaired reaches may be available on the same stream. In other cases, management activities could be modified to improve conditions at the sampling site.

Plains Region. Natural conditions apparently account for most stream impairment in the plains region. The potential for these streams to support healthy and diverse aquatic communities is naturally limited. Natural and human-induced factors such as dewatering, upland and streambank erosion, silty substrate,

high water temperatures, salinity, and poor streamside vegetation clearly limit the ability of these streams to support aquatic life.

Naturally limiting conditions also make these streams particularly susceptible to management activities. There is no doubt that grazing and other management activities aggravate naturally poor conditions, but it is difficult to determine the relative degree to which management and natural factors influence stream condition.

Although the assessments indicate a high incidence of impairment in this region, finding better (i.e., less impaired) reference sites would be difficult, if not impossible. An exception might be to find streams that had no influence from management activity. But given the prevalence of agriculture in this part of Montana, it is doubtful that a "pristine" plains stream exists. This, combined with the natural impairment, suggests that the chosen reference streams are at or near the potential for this region.

Although the plains streams certainly support aquatic organisms, their greatest potential value may be for wildlife habitat. This habitat is provided by the streams' riparian vegetation. Modification of management activities would have great potential for improving riparian vegetation and wildlife habitat. Side benefits would be improved aquatic habitat and streamflows.

PERIPHYTON

The Flora

Results of periphyton samples analyses, including QA/QC, are presented in Appendices 8, 9, and 10. Diatoms (Division Bacillariophyta), green algae (Division Chlorophyta), and blue-green algae or cyanobacteria (Division Cyanophyta) dominated the microflora of Montana reference streams. Representatives of all three divisions were found in all but one stream: green algae were missing from the East Fork Bull River sample. Other divisions represented were Chrysophyta (yellow-green or golden brown algae), Rhodophyta (red algae), and Euglenophyta (euglenoid algae). Mosses (Bryophyta) were recorded in samples collected from all but on of the West Side streams in the Northern Rockies Ecoregion.

The cumulative weighted rank of non-diatom genera within each algal division was calculated for each ecoregion/subregion (Table 4). This figure gives a rough approximation of the relative biomass of each division of non-diatom algae. Blue-green algae dominated the non-diatom microflora of Northern Rockies reference streams and green algae dominated the non-diatom microflora of plains reference streams. The two groups were co-dominants in streams of the Middle Rockies and the Montana Valley and Foothill Prairies. Red algae and golden-brown algae were subdominants in mountain streams, but they were rare or absent in samples from plains streams.

Dominance by blue-green algae may be a function of the relatively small inorganic nitrogen values in streams of the Northern Rockies Ecoregion (Figure 12). Blue-greens have a competitive advantage over other algae in such streams by being able to "fix" atmospheric or molecular nitrogen when bioavailable nitrogen in ionic form (nitrate and ammonia) is in short supply. Larger concentrations of nitrogen, such as those present in plains reference streams, tend to favor green algae.

Achnanthes minutissima was frequently the dominant diatom species in mountain and foothill streams. Several species in the genera Achnanthes, Cyclotella, Diploneis, Epithemia, Fragilaria, and Nitzschia dominated the diatom associations of plains streams. Although dominant diatom species varied considerably from stream to stream within an ecoregion, there was more consistency at the genus level (Appendix 11). On the basis of cumulative number of cells in each genus across all stands (streams) in an ecoregion, Achnanthes is the dominant diatom genus in mountain and most foothill streams and Nitzschia is dominant in plains streams.

Periphyton community types of Montana ecoregions based on the dominant division(s) of non-diatom algae (by volume) and the dominant genus of diatom (by cell numbers) are suggested in Table 5. By this scheme, reference streams in the Northern Rockies Ecoregion support Cyanophyta-*Achnanthes*

communities and reference streams in plains ecoregions support Chlorophyta-*Nitzschia* communities. Reference streams in the Middle Rockies Ecoregion and the Montana Valley and Foothill Prairies support an intermediate community type: Chlorophyta/Cyanophyta-*Achnanthes*. These community types, and subtypes of these communities in each of the ecoregions, may be defined further on the basis of algal taxa that the streams in those ecoregions/subregions have in common (Appendix 12). Several of these "diatom taxa in common" were limited to a single ecoregion/subregion (e.g., *Melosira distans* in the Northern Rockies/West Side/Granitics subregion). Such "exclusives" exhibit a degree of fidelity to this community subtype (Hanson and Churchill 1961, Daubenmire 1968) and may be used as ecological markers.

Other "diatom taxa in common" were recorded in all stands (streams) from more than one ecoregion. The diatom species *Cymbella minuta* was the only one counted in all periphyton samples collected from reference streams in mountain and foothill ecoregions, except spring creeks in the Montana Valley and Foothill Prairies. The diatom species *Navicula veneta* was the only one counted in all periphyton samples collected from plains reference streams. These cosmopolitan species here demonstrate a high degree of constancy within their respective community types.

Metrics

Mean, minimum, and maximum values for periphyton metrics in each ecoregion/subregion are presented in Figures 14 through 22. All but one of the metrics defines features of the diatom portion of the periphyton community. The one exception is the number of common non-diatom genera. (Here "common" means genera whose cells averaged more than one per field of view at 200X in a scan of a composite wet mount made from the original sample.)

Metrics were calculated in three categories. The rationale for these metrics is explained in the chapter entitled "STUDY DESIGN":

- 1. Periphyton Community Structure
 - Number Common Non-Diatom Genera
 - Number Diatom Species Counted ("Species Richness")
 - Percent Relative Abundance (PRA) Dominant Diatom Taxon
 - Diatom Species Diversity (Shannon)
- 2. Lange-Bertalot Pollution Tolerance Groups (Diatoms)
 - PRA Most Tolerant Group (Group #1)
 - PRA Less Tolerant Group (Group #2)
 - PRA Sensitive Group (Group #3)
- 3. Diatom Species Indicators
 - Number of Navicula plus Nitzschia Species Counted
 - PRA *Navicula* plus *Nitzschia* Species
- 1. Periphyton Community Structure. On average, significantly fewer common non-diatom genera were recorded in streams of the mountain and foothill ecoregions than in streams of the plains ecoregions (Figure 14). One notable exception was the Big Hole River, which had 15 common non-diatom genera. The site sampled on the Big Hole (Fishtrap Fishing Access) was later found to be influenced by agricultural practices upstream and for this reason should not be used for reference purposes.

Besides having fewer non-diatom genera, streams of the mountain and foothill ecoregions generally supported fewer diatom species than streams in the plains ecoregions (Figure 15). However, streams in the West Side/Granitics subregion of the Northern Rockies Ecoregion appeared to support a significantly larger number of diatom species than streams in other mountain ecoregions and subregions.

The smaller number of algal genera and species in mountain species is probably a function of the more severe natural conditions in these streams. Mountain streams tend to be steeper, faster, colder, darker, and poorer in nutrients compared to plains streams. These harsh conditions may increase competition for resources and limit the number of algal niches available.

The percent relative abundance (PRA) of the dominant diatom species generally ranged between 20 and 40 (Figure 16). This metric tended to be somewhat higher in the Northern Rockies Ecoregion. This may also be explained by the more austere natural conditions of streams in this ecoregion. The higher values for the "special creeks" are probably due to human-caused disturbances, which make these streams less than ideal for reference purposes.

Diatom species diversity (Shannon) followed the same pattern as number of diatom species counted (Figure 17). The very low Shannon diversity (2016) recorded for the East Fork Bull River probably reflects the especially cold, nutrient-poor, and light-poor condition of this stream. Again, the lower diversity values for the "special creeks" are probably due to human-caused pollution.

Periphyton community structure metrics were also calculated for samples collected in prior years from least-impaired reference streams in the West Side/Non-Granitics subregion of the Northern Rockies, the Northwestern Glaciated Plains, and the Northwestern Great Plains (Appendix 13). Metric values calculated for these earlier samples are generally comparable to the ones calculated for the 1990 samples collected from the same ecoregions/subregions.

2. Lange-Bertalot Pollution Tolerance Groups. The cumulative PRA of diatoms in the most tolerant group (#1) averaged less than 7 in streams of the mountain and foothill ecoregions and between 8 and 14 in the plains ecoregions (Figure 18). The cumulative PRA of diatom species in the less tolerant group (#2) averaged less than 40 in streams of the mountain and foothill ecoregions and 50 or more in the plains ecoregions (Figure 19). Conversely, the cumulative PRA of diatom species in the sensitive group (#3) averaged 60 or more in the mountain and foothill ecoregions and less than 50 in the plains ecoregions (Figure 20).

To a great extent, these basic differences in the relative abundance of pollution-sensitive and pollution-tolerant diatoms between the mountain and plains ecoregions reflect basic differences in the natural levels of such variables as temperature, nutrients, sediments, and dissolved salts. However, all but two of the plains reference streams exhibit some level of impairment (Table 3), so human disturbance also accounts for some of these differences.

The PRA values for diatoms in the three pollution-tolerance groups can be combined into a single pollution index as follows. First, the decimal fraction of diatoms in each group is multiplied by its group number. (For example, if half the diatoms in a sample belong to the sensitive group (Group #3), you would multiply 0.50 by 3 to get 1.50.) Then the products for the three groups are added to yield the pollution index. This index will range from 1.00 (very polluted) to 3.00 (unpolluted).

3. Diatom Species Indicators. Both the cumulative PRA of species and number of species counted in the genera *Navicula* and *Nitzschia* were significantly larger in the plains ecoregions than in the mountain ecoregions, with intermediate values in the Other Geology subregion of the Middle Rockies Ecoregion and in the Montana Valley and Foothill Prairies (Figure 21 and 22). These motile diatoms are adapted to maintaining their position on unstable substrates and appear to be useful as indicators of sedimentation, based in part on these ecoregional differences.

Although not common in any of the reference streams, species of the motile genus *Surirella* often account for a large percentage of the diatom cells in Montana streams that have serious sedimentation

problems, for example lower Prickly Pear Creek near Helena. For this reason, PRA *Surirella* spp. Should be included in this metric when comparing an impaired stream to a reference stream or a control site.

Ecoregion Groups

A look at Figures 14 through 22 reveals some distinct differences between mountain and plains streams for several of the periphyton metrics, and a degree of uniformity of values within each of these regions. On this basis, we propose mountains and plains as the most practical geographical units for summarizing periphyton metric data (Table 6) and for developing biological criteria for periphyton.

Values for periphyton metrics from foothill streams were mostly intermediate between values for mountain and plains streams, but generally much closer to values for the mountain streams (Figure 145 through 22). This was also true for streamflow and several water quality constituents (figure 2 through 13). Since most of the foothill streams and few of the mountain streams were impaired, the small differences between these two sets of metrics may be due to different levels of impairment rather than to natural factors.

We believe that metric values for unimpaired mountain streams represent the **potential** for periphyton metrics for streams in the Montana Valley and Foothill Prairies Ecoregion. For this reason, and because the foothill streams suffered only minor impairment, we combined the mountain and foothill data in calculating summary metric values (Table 6).

MACROINVERTEBRATES

Results of macroinvertebrate sample analyses are presented in Appendix 14. Much of the following discussion is based on the macroinvertebrate contractor's report (Wisseman undated) and supplemental documents.

Ecoregion Groups

DECORANA produced three major groups of reference streams corresponding to the three major geographical regions of Montana-mountain, plains, and foothills (Figure 23). These regions consolidate the ecoregions proposed by Omernik and Gallant (1987), as follows:

Mountains: Sites in both the Northern Rockies and Middle Rockies Ecoregions exhibited a relatively tight clustering, especially on axis one.

Foothills: Invertebrate communities from the Montana Valley and Foothill Prairies Ecoregion clustered together, including all spring creeks except for Big Spring Creek

Plains: Sites from all three plains ecoregions in Montana exhibited a tight clustering

Ordination analyses such as DECORANA allow for more consistent interpretation and evaluation of large, complex data sets. DECORANA analyzes complex data in multiple dimensions. The results of these analyses are usually summarized on 2 or 3 axes, which explain most of the meaningful variability (Gauch 1982). Only the first two axes have been used in this analysis. Variance accounted for by the axes were 0.867 for axis 1 and 0.565 for axis 2.

The sites are replotted in Figure 24 to highlight mountain, plains, and foothill stream groups. Axis 1 produces a strong division between plains and mountain streams, with foothill streams intermediate and contiguous with the mountain streams. Axis 1 appears to be related to physical features such as water temperature, gradient, water chemistry, and substrate. Axis 2 may be related to discharge since the sequence of mountain sites along the axis is roughly in order of increasing discharge.

DECORANA by itself does not explain the causes of the site positionings. Because of the large number of variables involved, further explanation of the causes of the observed patterns would require larger and more focused data sets.

The foothills group contains mostly Montana Valley and Foothill Prairie sites. They are grounded tightly on axis 1, but spread out on axis 2. Except for Big Spring Creek, this group was tighter than mountain streams.

Big Spring Creek is an outlier. This is an unusual stream and may not be an appropriate ecoregion reference. Big Spring Creek is a large creek (estimated flow: 120cfs) fed by a large spring. It is located in central Montana in the Northwestern Great Plains Ecoregion. A fish hatchery at the source of the creek is located just upstream of the sampling station and contributes organic matter and nutrients to the stream.

Expansion of the mountain portion of the DECORANA plot (Figure 25) produced no ecoregion subgroups, except for a possible affinity between the Northern Rockies/West Side Granitics and the Middle Rockies/Other Geology subregions. This observation may warrant further exploration, however we feel that the existing data are insufficient to draw firm conclusions.

Expansion of the Plains portion of the DECORANA plot (Figure 26) produced no obvious ecoregion subgroups.

We ended the multivariate analysis at this point. The initial DECORANA produced clear results and allowed us to establish regional groups of streams. However, we do not feel that the 1990 data set is robust enough to warrant continued manipulation in an attempt to further subdivide these groups. The sample size taken from each ecoregion and major habitat type (e.g., small, medium, and large streams) is too small to support further analysis.

In this study and in a similar study of least-impaired streams in eight Oregon ecoregions (Whittier et al. 1988), the most appropriate initial partitioning of macroinvertebrate communities was into mountain and lowland streams. Though fish and periphyton communities displayed some tendency to group by ecoregion in the Oregon study, lotic invertebrate communities in western North America do not seem to cluster into the ecoregions defined by Omernik and Gallant (1987). It may be more productive to further divide the mountain, plains, and foothills regions according to size of the stream or the size of the drainage area, rather than into smaller geographic units.

The Fauna

Mayflies, stoneflies and caddisflies- insects belonging to the orders Ephemeroptera, Plecoptera, and Trichoptera- dominated **mountain** stream macroinvertebrate communities. Respectively, they contributed an average 51, 9, and 10% of the organisms in each community. The six most dominant taxa were mayflies (Table 7). *Baetis tricaudatus*, an ubiquitous and relatively pollution-tolerant species, ranked first.

The remaining dominant taxa in **mountain** streams were a mixture of highly intolerant to moderately tolerant forms. Chironomid midges averaged 6% of the mountain stream communities, however they may have been under-represented in this study due to the relatively large mesh of the sampling nets used. Seven midge genera per stream was the average. Non-insect taxa averaged only 2 at each site, and accounted for only 1% of the organisms. Odonata, Hemiptera, Megaloptera, and Lepidoptera were absent.

The invertebrate fauna of the **foothills** streams was most similar to the mountain stream fauna, and intermediate between plains and mountain faunas. On average, caddisflies accounted for about 25% of the organisms, chironomids 18%, beetles 15%, mayflies 15%, and true flies (dipterans) other than chironomids 3%. Caddisfiles such as *Lepidostoma* and *Brachycentrus* were often abundant.

Benthic invertebrate communities of **plains** streams contrasted sharply with those from mountain streams. Non-insects, water boatmen, damselflies, chironomid midges, blackflies and mayflies were the most abundant groups. *Physella*, a highly tolerant snail, was the dominant taxon, followed by *Hyallela azteca*, a tolerant amphipod crustacean, and *Caenis*, a silt-tolerant mayfly. Each taxon represented an average of 7% to 8% of the organisms in each sample. Most of the abundant taxa can be classes as highly tolerant of organic and nutrient pollution, high temperatures, depressed dissolved oxygen, and fine sediments.

Metrics

A first generation set of metrics was selected for Montana (see below). These need further evaluation as more data become available. In particular, macroinvertebrate data need to be examined and correlated with other measures of water and habitat quality from streams exhibiting a wide range and degree of impairment.

The metrics that were selected are:

- Taxa Richness (number of taxa)
- Number of Ephemeroptera, Plecoptera, and Trichoptera Taxa (EPT Taxa)
- % Chironomidae (% of organisms in family Chironomidae)
- % Collectors (% of organisms that are collectors)
- % Scrapers (% of organisms that are scrapers)
- Scrapers/Scrapers+Filter Feeders (Sc/Sc+FF) (number of organisms)
- % Dominant Taxon
- Hilsenhoff Biotic Index (HBI)
- Community Tolerance Quotient (CTQa)
- Quantitative Similarity Index for Taxa (QSI-Taxa)
- Quantitative Similarity Index for Functional Feeding Groups (QSI-FFG)
- Dominants-In-Common 5 (DIC-5)

When Rapid Bioassessment Protocol III is adapted for regional use, there is an opportunity to incorporate taxonomic metrics that are based on genus- or species-level identifications and the autecology of those genera and species. This approach employs individual taxa or assemblages of taxa that are ubiquitous within a region and are strongly correlated with water quality. It has been widely used in Europe (Hellawell 1986).

Regional means and coefficients of variation for the selected metrics are given in Table 8. Means and ranges of metrics values for each ecoregion/subregion are displayed in Figures 27 through 35. Discussion of individual metrics follows.

<u>Taxa Richness.</u> This metric is the total number of taxa that have been identified from a site. There is usually a direct relationship between taxa richness and water quality. As water quality declines so does taxa richness, and vice versa.

This metric must be interpreted with caution. Taxa richness, as well as EPT richness, is sensitive to different levels of effort in sampling and subsampling. A bias can be introduced into comparisons when relative sampling effort is not taken into account. Also, slight or moderate enrichment of water bodies can lead to an increase in taxa richness, especially in streams that are naturally very unproductive. Further enrichment will eventually cause most sensitive forms to disappear.

Taxa richness may also increase from headwaters to mouth, even in the face of declining water quality. Intermediate sites in a watershed may have natural communities composed of both headwater and riverine taxa, generating taxa richness values that are larger than those for upstream or downstream communities. This may occur even if habitat and water quality decline somewhat in an upstream to downstream direction. Moderate or severe impairment usually accelerates the impoverishment of fauna along a longitudinal gradient.

Mountain streams tended to support a larger number of taxa than plains stream (Figure 27). The average number of taxa for mountain, foothill and plains streams was 34, 34, and 29, respectively.

EPT Richness. Ephemeroptera, Plecoptera, and Trichoptera (mayflies, stoneflies, and caddisflies) are orders of aquatic insects that are generally intolerant of pollution, although individual EPT taxa display a wide range of pollution tolerance. Mayflies and caddisflies are found in a wide range of aquatic habitats. Stoneflies are generally restricted to cooler, cleaner, running water. In healthy running water habitats, most of the taxa belong to these three orders.

There were significantly more EPT taxa in the mountain streams than there were in the plains streams (Figure 28). The average number of EPT taxa for mountain, foothill and plains streams was 22,16, and 6, respectively.

<u>% Chironomidae.</u> The family Chironomidae (midges) has been highly successful in freshwater and accounts for more than half of all macroinvertebrate species in many aquatic habitats (Bode, 1990). Diverse assemblages of midge species can be found in all but the most severely polluted waterbodies. Midges are common in most running waters.

Disproportionate dominance by chironomid midges often indicated impairment. Waterbodies severely stressed by pollution (nutrients, sediment or toxins) are often dominated by members of this family of true flies.

Midges were more common in plains streams than they were in most mountain streams (Figure 29). Mountain reference streams averaged 7 genera of midges, and they accounted for 9% of all organisms. Foothill streams averaged 18% midges in 9 genera, while plains streams had 23% midges in 9 genera.

<u>% Collectors.</u> Collectors are aquatic insects that feed on fine sediment enriched with particles of organic matter. The proportion of the invertebrate community composed of collectors can be a useful indicator of habitat and water quality. If collectors are increasing at a site, a declining trend in habitat and/or water quality is indicated.

Collectors generally increase in abundance in downstream direction. They also exhibit and inverse relationship with stream gradient. As stream gradient decreases, fine sediment settles to the bottom and collectors increase. Slight or moderate nutrient enrichment can cause an increase in the abundance of collectors in moderate or low gradient streams.

The relative abundance of collectors was the most uniform of all functional feeding group metrics in this study. For example, the coefficients of variation (CV) for percent collectors in foothills (0.47), plains (0.13), and mountain (0.16) regions were much smaller than those for filter feeders (CVs all 1.4). For statewide use, collectors were therefore considered a better habitat and water quality indicator than other functional feeding groups.

Collectors were generally more abundant in plains streams than in mountain streams (Figure 30).

<u>% Scrapers.</u> Insects in the scraper feeding group feed by grazing periphyton from rocks and other hard surfaces found on the bottom of streams. A diverse and abundant scraper community reflects good habitat and water quality.

Nutrient enrichment usually reduces the percentage of scrapers in a community. This is because preferred diatom dominated periphyton communities are replaces by unpalatable filamentous algae and bacterial mats.

Large amounts of fine sediment can settle on hard surfaces during periods of low flow, smothering periphyton and inhibiting scrapers.

Excessive scouring of streambed substrates by large amounts of silt and sand transported during high flows can remove the scrapers' periphyton food base.

Scrapers were generally much more abundant in mountain streams that in plains streams (Figure 31). The average percentage of scrapers was 36 in mountain streams, 6 in plains streams, and 24 in foothill streams. This metric is not well suited for use in the plains ecoregions because of the very low percentage of scrapers here.

<u>Scrapers/Scrapers+Filter Feeders.</u> This metric is the ratio of the number of organisms in the group generally indicative of high quality habitat and water (scrapers) to the number of organisms in the group indicative of lower quality (filter feeders). Larger values indicate increasing dominance by scrapers and better habitat and water quality.

Values for this metric spanned the full possible range (Figure 32), with lower values for plains streams (low scraper abundance) and higher values for mountain streams (low filter feeder abundance). Low scraper abundance in the plains streams limits the utility of this metric in this region. On the other hand, the small numbers of filter feeders in mountain streams may make this metric very sensitive to an increase in filter feeders and a decrease in scrapers resulting from organic enrichment.

% Dominant Taxon. This metric is the percent relative abundance of the most numerous taxon found in a community. It is a simple measure of diversity. Plafkin et al. (1989) suggested that the numerically dominant taxon in minimally impacted streams should account for less than 20% of the community. Values produced in this study were generally larger. This may be due to the coarse mesh (1mm) of the nets used, which would allow relatively small invertebrate taxa to escape, resulting in disproportionate representation by the dominant taxon. Using these nets, the criterion probably should be larger than 20%.

In general, the more stressed an aquatic ecosystem, the greater will be the percentage of individuals in the dominant taxon. The dominant taxon may account for over 90% of the individual in a severely stressed community.

The percent relative abundance of the dominant taxon was somewhat larger in plains streams than in mountain streams (Figure 33). Average values measured in this study were 29% in mountain streams, 35% in plains streams, and 35% in foothill streams. The plains streams may have large values because most of these ecosystems are stressed from fine sediment loads and (or) low flows.

<u>Hislenhoff Biotic Index.</u> The Hilsenhoff Biotic Index (HBI) (Hilsenhoff 1987) was designed as an index of organic enrichment to streams in the Midwest. It is also believed to be a good indicator of enrichment by inorganic nutrients.

The HBI is based on tolerance values assigned to aquatic insect taxa. It has been modified for family level effort (Hilsenhoff 1988) and to include non-insect macroinvertebrates (Plafkin et al. 1989). In this study we took advantage of both of these modifications as well as information available for Western taxa (Wisseman, unpublished data).

Possible values range from 0-10, with 0 indicating a community composed entirely of taxa sensitive to nutrient enrichment, and 10 indicating sewage lagoon conditions. HBI values for plains streams were all larger than 6, while values for mountain streams were all smaller than 4 (Figure 34).

Like taxa richness and % collectors, HBI values will generally increase along a longitudinal gradient, even in pristine streams. This reflects a natural deposition and accumulation of nutrients in lower gradient habitats. Also, prairie streams generally have higher HBI values than forested streams. Lower gradient, higher water temperature and more sunlight lead to more plant production, higher decomposition rates, and lower dissolved oxygen levels.

<u>Community Tolerance Quotient (CTQa).</u> This metric is similar to the Hilsenhoff Biotic Index. The CTQa was developed for use in Western streams to assess nonpoint source pollution (USFS 1989). Patterns for the reference stream data are similar to those for the HBI (Figure 35).

Quantitative Similarity Index for Taxa (QSI-Taxa). The QSI-Taxa was developed by Shackleford 1988. This index compares the similarity of two communities, both in terms of taxa present and their relative abundance. Values for this index range from 0 to 1, with identical communities having a value of 1 and totally dissimilar communities having a value of 0.

Ordinarily a comparison would be made between a reference community and an impaired community, but in this study each reference community was compared to a composite reference community for that ecoregion or group of ecoregions. Average QSI-Taxa values for these comparisons were 0.38 for mountain streams, 0.35 for plains streams, and 0.29 for foothill streams (Table 8).

<u>Quantitative Similarity Index for Funcional Feeding Groups (QSI-FFG)</u>. The QSI-FFG is calculated the same was as the QSI-Taxa, except that it used the relative abundance of functional feeding groups instead of the relative abundance of taxa. It is designed to compare the functional integrity of two communities.

Average values for Montana reference streams compared to regional composite communities were 0.73 for mountain streams, 0.68 for plains streams, and 0.61 for foothill streams (Table 8).

Macroinvertebrate Habitat

Table 9 summarizes macroinvertebrate habitat assessments conducted at each reference stream site in 1990. There was no obvious correlation between invertebrate community metrics and habitat scores (Figures 36 through 41). There appears to be weak correlation between habitat score and taxa richness (Figure 36), EPT richness (Figure 37), and percent dominant taxon (Figure 40).

This survey included only minimally-impaired reference streams. Therefore, only a narrow range of possible habitat assessment and invertebrate community metric values is represented. A much wider range of values will be needed to properly test correlations between habitat quality and invertebrate metric values.

The macroinvertebrate habitat assessments used for this study were a weak predictor of macroinvertebrate community health. We feel that the Habitat Assessment Protocol proposed as a component of the RBP III (Plafkin et al. 1989) needs major modification to better quantify the relationship between macroinvertebrate habitat and macroinvertebrate community metrics.

FISH

Fish data retrieved from the Interagency Stream Fishery Database are presented in Appendix 15.

Reference streams in the mountain and foothill ecoregions supported cold-water species. Reference streams in the plains ecoregions supported cool-water and warm-water species.

Common fish species occurring in all reference streams within each ecoregion/subregion are listed in Table 10. The lists of species in common are identical for the spring creeks and "special creeks" of the foothill region. The lists are unique for the subregions of the Northern Rockies and Middle Rockies Ecoregions. The flathead minnow, lake chub, and longnose dace were common in all reference streams in the plains region.

Plains reference streams averaged more native and introduced species of fish than mountain or foothill reference streams (Figures 42 and 43). Mountain and foothill streams generally supported fewer than 10 species of native and non-native fish, while plains streams tended to support more than 10 fish species (Figure 44).

As with algae, the lower fish diversity in mountain streams may reflect the more austere habitats provided by these ecosystems. These differences may also be due to the different origins and biogeographic histories of Montana's warm water and cold water fish faunas (Frank McCormick, pers. Comm., 14 February 1992).

CONCLUSIONS AND RECOMMENDATIONS

WATER QUALITY

- Based on streamflow and water quality, reference streams tended to cluster into a
 mountains group and a plains group. Streams of the Montana Valley and Foothill Prairies had
 intermediate values, which, for most parameters, were closer to those recorded for mountain
 streams.
- 2. Mountain and foothill reference streams tended to have more flow than plains streams. Continuing drought on the plains no doubt enhanced this differential. Another reason is because few large streams originate within the plains region; most large streams flowing through the plains originate in adjacent mountain ecoregions. These streams were avoided in selecting plains reference streams because they are impaired and not typical of streams in this region.
- 3. Streams in the mountain region and unimpaired streams in the foothill region had temperatures that support cold-water aquatic life. Plains streams had temperatures that support warm-water aquatic life.
- 4. Mountain and foothill streams were freshwater streams; plains streams were either freshwater or brackish. Mountain and foothill streams had calcium bicarbonate waters; plains streams had sodium sulfate or sodium bicarbonate waters.
- 5. Measured total suspended sediment values afford a "high level of protection" to aquatic life in mountain and foothill streams, and a "high" to "low" level of protection in plains streams.
- 6. All Montana reference streams were basic, that is, all had pH values larger than 7.00 standard units. All mountain and foothill streams had total alkalinity values smaller than 200 Mg/L CaCO₃; all but one plains stream had total alkalinity values larger than this amount. Total hardness rating for mountain and foothill streams were "soft" to "hard"; plains streams were "moderately hard" to "very hard" (see Figure 8).
- 7. Mountain streams and spring creeks tended to be phosphorus limited; plains streams tended to be nitrogen limited. The Northern Montana Glaciated Plains had the largest nutrient values of any ecoregion. Nuisance algae growths are most likely in streams of this ecoregion and in the Montana Valley and Foothill Prairies.
- 8. Water quality on STORET should be evaluated to determine if there is a water quality basis for the ecoregions of Omernik and Gallant (1987) and the subregions identified in this report. If the ecoregions and subregions do have distinctive water quality "signatures", then water quality differences between them will be larger than the water quality differences within them. If not, then consolidating ecoregions into mountain and plains regions, as proposed here is valid.

STREAM REACH ASSESSMENTS

 In several streams, the sample sites were **not** located on the reach that was assessed as least impaired (Table 11). For these stream, sample sites should be relocated to the least impaired reach recommended in Table 11. The highest priority for changing sample sites should be given to those streams with the most disparity in impairment ratings between existing and recommended locations.

- Despite frequent impairment, the plains reference sites probably represent the potential of
 this region for supporting aquatic uses. Natural limitations combined with a prevalence of
 agriculture will make it difficult to find better or less impaired reference sites. However, we
 will continue to look for less-impaired reference streams in the plains ecoregions.
- 3. In the mountain region, channel types and land used often differ between upstream and downstream reaches. These differences may significantly influence channel condition and aquatic use support and should be further investigated. If the differences are meaningful, ecoregion or subregion boundaries should be modified.
- 4. The Montana NPS Stream Reach Assessment is a rapid method for providing useful information relative to general steam conditions and factors influencing stream conditions. However, the information provided is limited because it is based on minimal, subjective field observation. If long term reference sites are established to define benchmark biological criteria, then comprehensive upstream surveys of all potential or existing stream influences should be completed.
- 5. Streams in foothill region have a good potential to fully support aquatic uses through improved land management practices. The reference reaches exhibit relatively minor impairment, mostly due to management activity rather than from natural factors. Better reference sites may be available in this region because management activities dominate the land. Since alternative sites are limited, management modification on existing reference steams should be encouraged in order to bring reference sites to their full potential.

PERIPHYTON

- Reference streams in the Northern Rockies Ecoregion support Cyanophyta-Achnanthes
 periphyton communities and reference streams in the plains region support ChlorophytaNitzschia communities. Reference streams in the Middle Rockies Ecoregion and the Montana
 Valley and Foothill Prairies support an intermediate type: Chlorophyta/CyanophytaAchanathes.
- 2. Genus and species richness and diatom diversity tended to be smaller in the mountain and foothill regions than in the plains region; percent relative abundance of the dominant taxon tended to be larger in the mountain and foothill regions than in the plains region. This is probably a function of the more austere natural conditions in the mountain and foothill streams. These streams tend to be steeper, faster, colder, darker and poorer in dissolved ions and nutrients compared to plains streams. These harsh conditions may increase competition for resources and limit the number of algal niches available. Of the four metrics used, the Shannon diversity index appears to be the best overall indicator of periphyton community structure and natural or man-caused stress.
- 3. Plains streams tended to have more diatoms in the pollution tolerant groups and fewer diatoms in the pollution sensitive group than streams in the mountain and foothill regions. A combination of natural factors and man-caused impairment probably accounts for these differences. We recommend combining results for the three Lange-Bertalot pollution tolerance groups into a single pollution index as described elsewhere in this report.
- 4. Both percent relative abundance (PRA) and number of species counted in the motile diatom genera *Navicula* and *Nitzschia* appear to be potentially good indicators of sedimentation. Both PRA and number of species were significantly larger in plains streams than in mountain streams. We recommend using PRA as the more sensitive of the two metrics, and adding the PRA of *Surirella* species when this genus is present. We also recommend researching the relationships between this metric, stream gradient, and total suspended sediment. This could be accomplished by examining data from impaired streams

that have a range of gradients and sediment loads. The relationships should be established independently for streams in the mountain and plains regions.

5. This study does not indicate a partitioning of periphyton metrics by the ecoregions of Omernik and Gallant (1987). Periphyton data appear to partition best into mountain and plains regions, with foothill streams included in the mountain region. Metric values for unimpaired mountain streams probably represent the potential for periphyton metrics in foothill streams. To test these conclusions, periphyton data and supporting data should be collected from several more unimpaired streams in each of the ecoregions and subregions. Periphyton metrics should be compared to environmental variables using DECORANA. Until then, periphyton assessment protocols and biocriteria should be developed separately for plains streams and mountain streams, the latter including foothill streams.

MACROINVERTEBRATES

- Data from this study indicate that mountain stream macroinvertebrate communities are
 dominated by mayflies, stoneflies, and caddisflies. Plains stream communities were quite
 different, with non-insects, water boatmen, damselflies, midges, blackflies and mayflies
 dominant. Macroinvertebrate communities from foothill streams were intermediate but most
 similar to mountain communities. Foothill stream communities were dominated by
 caddisflies, midges, beetles, and mayflies.
- 2. Increase standardization of sampling effort. Complete standardization is impossible due to the physical differences between sites. However, limiting the duration of the kick sample to 1 minute may reduce sample variance and make it easier to compare data between sites.
- 3. Continue to use nets with 1mm mesh. Although this is a relatively coarse mesh, it is consistent with the **rapid** bioassessment approach used in this survey. A net with finer mesh would collect many immature macroinvertebrates. Identifying these is difficult and time consuming. Any mesh size represent a compromise between collection efficiency, sample processing efficiency, and cost. Nets with very fine mesh are inefficient collectors because of backwash, especially in rapidly flowing streams. The D-frame aquatic nets that we used (Wards No. 10 W 0620) are sturdy, relatively inexpensive, and readily available. They have a deep (18") bag that minimizes backwash. Continued use of these nets in surveys of impaired streams would allow for direct comparison of the data collected to reference stream data in this report.
- 4. DECORANA suggests that Montana macroinvertebrate communities partition best into mountain, plains and foothill communities, and not by ecoregion. The set of date we collected was limited relative to the population of streams in the state; conclusions drawn from these data should be viewed with caution. We recommend a more intensive follow-up effort on third-order mountain streams. The mountain ecoregions contain a large number of relatively unimpaired streams to choose from. We recommend sampling 5 streams from each of the Middle and Northern Rockies ecoregions to examine partitioning of community composition. If the ecoregions outlined by Omernik and Gallant (1987) are valid for macroinvertebrate communities, the differences between Northern Rockies and Middle Rockies communities will be larger than the differences between communities within an ecoregion. If not, the conclusions from this study are valid.
- 5. The Montana Valley and Foothill Prairies Ecoregion contains may of the state's most productive fishing streams and several streams with watershed improvement projects under the Section 319 Nonpoint Source Pollution Control Program. More intensive effort is recommended in this ecoregion to better define reference conditions and to relate metrics from impaired streams to kinds and degrees of management activity.

MACROINVERTEBRATE HABITAT

Scores derived from the macroinvertebrate habitat assessments (Plafkin et al. 1989) did not correlate well with values calculated for the macroinvertebrate metrics. This suggests the need for a better habitat assessment protocol. Until one is developed, use of the current protocol is till worthwhile for the sake of providing information that supports the macroinvertebrate data.

FISH

- 1. Reference streams in the mountain and foothill regions support cold-water fish species; reference streams in the plains region support cool-water and warm-water species.
- 2. There is not as much uniformity in the makeup of fish faunas among ecoregions and subregions of the mountain region as there is in the plains region. No species were found in all streams of the Northern Rockies and Middle Rockies Ecoregions. The fathead minnow, lake chub, and longnose dace were common in all reference streams in the plains region. The spring creeks and the special creeks in the foothill region had four species in common: brown trout, rainbow trout, mountain whitefish, and mottled sculpin.

Mountain and foothill streams have depauperate fish faunas compared to plains streams. Reference streams in the plains region generally supported more than 10 species of native and introduced fish, while reference streams in the mountain and foothill regions tended to support fewer than 10 fish species.